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# MICROBIAL GROWTH YIELD ESTIMATION FOR INCOMPLETE DEGRADATION AND MACRONUTRIENT UTILIZATION – THE CASE OF GLYPHOSATE

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Standardized simulation tests using isotope-labelled chemicals are carried out for persistency assessment. In these tests, formation of non-extractable residues (NER) is typically observed. Recent theoretical, experimental and analytical advances have helped deciphering the nature and composition of NER [1]. Among other types, biogenic NER is formed as the microorganisms use the test chemical as a source of carbon and energy, and methods for their prediction based on microbial yield estimations have been accordingly developed [2]. Evaluating and predicting biogenic NER formation still remains a challenge for test chemicals that (i) are utilized as a source of macronutrients (besides carbon); (ii) undergo incomplete degradation via multiple pathways, with formation of dead-end transformation products. In the present study, we addressed this challenge by (i) extending a recently developed microbial yield estimation method (MTB—Microbial Turnover to Biomass) by accounting for macronutrient limitation and incomplete degradation; and (ii) incorporating this method in a dynamic model to describe the fate of glyphosate and its main transformation product (AMPA) in a water-sediment simulation test. Glyphosate is known to be biodegraded via, at least, two pathways, one forming the recalcitrant metabolite AMPA [3]. For the assessment of model simulations, we used published data from turnover experiments using co-labelled <sup>13</sup>C<sub>3</sub><sup>15</sup>N-glyphosate [4]. Determination of microbial yield for different degradation pathways required consideration of the flow of elements, energy and electrons. By adapting three parameters, namely the number of electrons and the number of C or N atoms that can be acquired in a transformation step, the MTB method could be used to predict the growth yield when either N or C are limiting growth. The results showed that the formation of AMPA reduces the growth yield (taken as g bacteria per mol of glyphosate metabolized) by more than threefold, and that glyphosate is a better source of nitrogen than of carbon. Dynamic model simulations adequately described the degradation of glyphosate and the formation of CO<sub>2</sub> and AMPA in the water-sediment test. In particular, balancing <sup>13</sup>C and <sup>15</sup>N allowed discriminating which macronutrient (carbon or nitrogen) limited microbial growth. Accordingly, the switch in glyphosate transformation pathway from full mineralization to the formation of AMPA could be explained by an initial nitrogen deficit, which during the experiment changed to nitrogen saturation and demand for carbon, thus affecting the formation of biogenic NER. These findings highlight the benefit of combining advanced prediction methods and experimental approaches to obtain deeper insights into microbial metabolism, chemical persistence and biogenic NER formation.

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